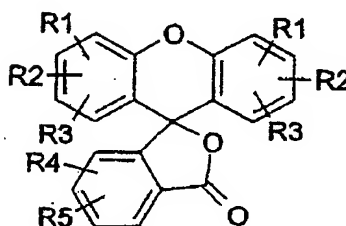


CLAIMS

1. A phthalein of general formula (I):



(I)

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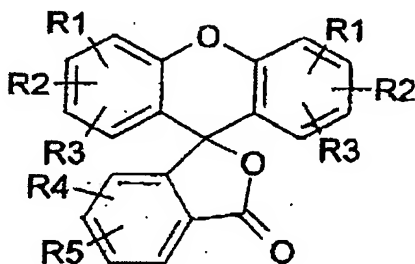
in which R1, R2, R3, R4 and R5, which are identical to or different from one another, are chosen from the group comprising the following radicals or groups: hydrogen, hydroxyl, halogen, acetyl, amino, phosphate, nitro, sulfonate, carboxyl, alkylcarboxyl having from 2 to 30 carbon atoms, alkyl having from 1 to 30 carbon atoms, cycloalkyl having from 3 to 12 carbon atoms, alkyloxy having from 1 to 30 carbon atoms, haloalkyl having from 1 to 30 carbon atoms, hydroxyalkyl having from 1 to 30 carbon atoms, alkyl ester having from 2 to 40 carbon atoms, nitroalkyl having from 1 to 30 carbon atoms, carboxyalkyl having from 2 to 30 carbon atoms, aminoalkyl having from 1 to 30 carbon atoms, sulfoalkyl having from 1 to 30 carbon atoms, aryl, aryloxy, arylalkyl, haloaryl, aryl ester, succinimidyl ester, isothiocyanate, maleimide, iodoacetamide, haloacetamide, chlorosulfonic, purine or pyrimidine bases, monosaccharides, preferably hexoses or pentoses, oligosides and polysides, polypeptides, proteins and phospholipids,

R1 and R5 are not hydrogen when R1 is a group $-\text{CH}_2-\text{CH}_2-\text{COOH}$, R2 is a hydroxyl group and R4 is a group $-\text{COOH}$, these phthaleins containing no more than 1% by weight, preferably no more than 0.5% by weight, and even more preferably not more than 0.2% by weight, of residual impurities.

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2. The phthalein as claimed in claim 1 consisting of fluorescein.

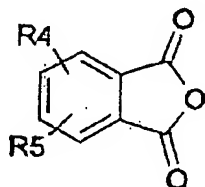
3. A method for preparing phthaleins, from which the residual impurities have been removed, having the general formula (I):



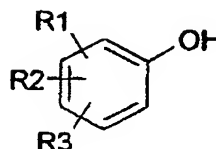
(I)

in which R1, R2, R3, R4 and R5, which are identical to
10 or different from one another, are chosen from the
group comprising the following radicals or groups:
hydrogen, hydroxyl, halogen, acetyl, amino, phosphate,
nitro, sulfonate, carboxyl, alkylcarboxyl having from 2
to 30 carbon atoms, alkyl having from 1 to 30 carbon
15 atoms, cycloalkyl having from 3 to 12 carbon atoms,
alkyloxy having from 1 to 30 carbon atoms, haloalkyl
having from 1 to 30 carbon atoms, hydroxyalkyl having
from 1 to 30 carbon atoms, alkyl ester having from 2 to
40 carbon atoms, nitroalkyl having from 1 to 30 carbon
20 atoms, carboxyalkyl having from 2 to 30 carbon atoms,
aminoalkyl having from 1 to 30 carbon atoms, sulfoalkyl
having from 1 to 30 carbon atoms, aryl, aryloxy,
arylalkyl, haloaryl, aryl ester, succinimidyl ester,
isothiocyanate, maleimide, iodoacetamide, haloacet-
25 amide, chlorosulfonic, purine or pyrimidine bases,
monosaccharides, preferably hexoses or pentoses,
oligosides and polyosides, polypeptides, proteins and
phospholipids,
R1 and R5 are not hydrogen when R1 is a
30 group -CH₂-CH₂-COOH, R2 is a hydroxyl group and R4 is a
group -COOH,

characterized in that a phthalic anhydride derivative of formula (II) is condensed with a phenol or naphthol compound of formula (III)



(I)



(III)

in which R1, R2, R3, R4 and R5 have the same meanings as above,
the condensation being carried out in a solvent consisting of an organic acid ester.

4. The method as claimed in claim 3, in which the compound of formula (III) is chosen from the group comprising resorcinol, orcinol, naphthol, pyrogallol, alkylaminophenol and arylaminophenol.

5. The method as claimed in either of claims 3 and 4, in which the solvent is an organic acid ester of formula (IV)



in which R₆ is chosen from the group comprising the following radicals or groups: hydrogen, alkyl having from 1 to 30 carbon atoms, cycloalkyl having from 3 to 12 carbon atoms, haloalkyl having from 1 to 30 carbon atoms, hydroxyalkyl having from 1 to 30 carbon atoms, nitroalkyl having from 1 to 30 carbon atoms, aryl, aryloxy, alkylaryl, arylalkyl, substituted arylalkyl, haloaryl, aryl ester, alkyl ester having from 2 to 40 carbon atoms, and alkyloxy having from 1 to 30 carbon atoms, R₇ representing one of the following groups: alkyl having from 1 to 30 carbon atoms, cycloalkyl having from 3 to 12 carbon atoms, haloalkyl having from

1 to 30 carbon atoms, hydroxyalkyl having from 1 to 30 carbon atoms, nitroalkyl having from 1 to 30 carbon atoms, aryl, aryloxy, alkylaryl, arylalkyl, substituted arylalkyl, haloaryl, aryl ester, alkyl ester having
5 from 2 to 40 carbon atoms, or alkyloxy having from 1 to 30 carbon atoms.

6. The method as claimed in one of claims 3 to 5, characterized in that the organic acid ester is chosen
10 from the group comprising methyl, ethyl, propyl or butyl benzoate, methyl, ethyl, propyl or butyl heptanoate, methyl, ethyl, propyl or butyl octanoate, methyl, ethyl, propyl or butyl laurate, methyl, ethyl, propyl or butyl myristate or methyl, ethyl, propyl or
15 butyl palmitate, and mixtures thereof.

7. The method as claimed in one of claims 3 to 6, characterized in that the condensation reaction is carried out at between 150°C and 250°C, optionally
20 under pressure.

8. The method as claimed in one of claims 3 to 7, characterized in that it is carried out in the presence of a catalyst chosen from the group comprising in
25 particular Lewis acids, such as ZnCl_2 or AlCl_3 , Brönsted acids such as H_2SO_4 or polyphosphoric acid, preferably an alkali metal hydrogen sulfate, and more preferably potassium hydrogen sulfate (KHSO_4) or sodium hydrogen sulfate (NaHSO_4).

30

9. The method as claimed in one of claims 3 to 8, characterized in that it comprises a step consisting in acidifying the product resulting from the condensation, in an anhydrous organic medium, by the addition of a
35 strong acid or one of its precursors, chosen from the group comprising sulfuric acid, hydrochloric acid, hydrobromic acid, hydrofluoric acid, hydriodic acid, polyphosphoric acid, pyrophosphate (P_2O_5), and mixtures thereof, the acidification being carried out until the

phthalein crystals resulting from the condensation are converted to phthalein crystals having a different structure.

5 10. The method as claimed in claim 9, characterized in that it comprises a step consisting in washing the product obtained after acidification, said washing step being carried out with a washing solution chosen from the group comprising water, alcohols, ketones, ethers
10 and halogenated solvents, pure or as a mixture.

11. A method for preparing a fluorescein having a purity such that its content of each of the by-products of the reaction is less than or equal to 0.2%, and
15 preferably less than or equal to 0.1%, the sum of the contents of each of these by-products being less than or equal to 0.5%, said method comprising the following successive steps:

- condensing phthalic anhydride with resorcinol, in a
20 solvent consisting of an ester of an aliphatic or aromatic organic acid, preferably ethyl or methyl benzoate or ethyl or methyl palmitate, in the presence of a catalyst chosen from the group comprising in particular Lewis acids or Brönsted
25 acids, and preferably an alkali metal hydrogen sulfate,
- suspending the red-colored crystals obtained in the preceding step in an anhydrous solvent chosen from the group comprising alcohols such as absolute
30 ethanol, ketones such as acetone, ethers, halogenated solvents, or mixtures thereof,
- acidifying the suspension thus obtained by the addition of a strong acid or one of its precursors, chosen from the group comprising in particular
35 sulfuric acid, hydrochloric acid, hydrobromic acid, hydrofluoric acid, hydriodic acid, polyphosphoric acid, pyrophosphate (P_2O_5), and mixtures thereof, until the red-colored crystals are converted to yellow-colored crystals exhibiting the X-ray

diffraction analysis of figure 2,

- washing the crystals obtained with a washing solution chosen from the group comprising water, alcohols, ketones, ethers and halogenated solvents, pure or as a mixture, this washing being continued until the yellow-colored crystals are reconverted to red crystals.

12. The method as claimed in one of claims 9 to 11, characterized in that the acidification is carried out by sparging gaseous hydrochloric acid into the phthalein suspension or by the action, on this phthalein, of hydrochloric acid in solution in the anhydrous organic solvent, preferably an alcohol, a ketone, an ether or a halogenated solvent, used alone or as a mixture, even more preferably isopropanol, absolute ethanol or acetone, pure or as a mixture.

13. The method as claimed in one of claims 3 to 12, characterized in that the catalyst used for the condensation reaction consists of the hydrogen sulfate of an alkali metal, preferably potassium hydrogen sulfate or sodium hydrogen sulfate.

14. A yellow-colored fluorescein crystal having the X-ray diffraction analysis of figure 2.

15. A yellow-colored 4',5'-dimethylfluorescein crystal having the X-ray diffraction analysis of figure 4.

16. A reddish-brown- or mahogany-colored 4',5'-dihydroxyfluorescein crystal having the X-ray diffraction analysis of figure 6.

17. A phthalein obtained by means of the method as claimed in any one of claims 3 to 13.

18. A fluorescein obtained by means of a method as claimed in any one of claims 3 to 13.

19. A 4',5'-dimethylfluorescein obtained by means of a method as claimed in any one of claims 3 to 13.

20. A 4',5'-dihydroxyfluorescein obtained by means of
5 a method as claimed in any one of claims 3 to 13.

21. The use of the fluorescein as claimed in claim 2
or obtained according to the method of claims 3 to 13,
in pharmaceutical applications in diagnosis, especially
10 in medical imaging, or in the field of biotechnological
applications.